

Adventitious rhizogenesis in *Bambusa nutans* and *Bambusa tulda*: Influence of seasonal variation, IBA and cutting type

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Abstract: The influence of seasonal variation, indole-3-butyric acid (IBA) and type of cuttings was examined on induction and growth of adventitious roots in *Bambusa nutans* Wall. and *Bambusa tulda* Roxb. Single-node culm and culm-branch cuttings from the mature culms were provided with immersion treatment for 24 h of either water (control) or 2 mM IBA in four different seasons, i.e., spring (mid February), summer (mid May), rainy (mid July), and winter (mid November) and maintained for two months in the mist chamber at the relative humidity of (70±5)% and the temperature of (30±2)°C. In *B. nutans*, adventitious rooting occurred in both types of cuttings in all the seasons with the best rooting in the summer season i.e., May (88% in culm cuttings) and the least in winter. On the contrary, adventitious rooting was recorded only in culm cuttings in spring and summer season in *B. tulda*. IBA treatment significantly enhanced rooting, root number and root length; registering 14 to 17 times improvement over control in the best rooting season. Three factor- interactions (season × cutting type × IBA treatment) were significant for rooting in *B. nutans* and all characteristics, except sprouting in *B. tulda*. Thus, single-node culm and culm-branch cuttings in *B. nutans* and culm cuttings in *B. tulda* treated with 2 mM IBA during spring (February) to summer (May) season are recommended for their clonal multiplication.

Keywords: *Bambusa nutans*; *Bambusa tulda*; branch cuttings; culm cuttings; rooting; sprouting

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Introduction

Over exploitation due to ever-increasing population pressure and new demands for industrial uses, especially pulp and paper, have resulted in enormous reduction of bamboo stocks, which needs to be amended by large-scale plantations of versatile and rapidly growing bamboo species like *Bambusa nutans* Wall. and *Bambusa tulda* Roxb. *B. nutans* occurs naturally in sub-Himalayan tracts but is also cultivated in north-west India, Orissa and West Bengal, while *B. tulda* is a native to north and north-east India, Bangladesh, Myanmar and Thailand (Gamble 1896). The strong and straight culms of *B. nutans* are used variously, mainly as poles and paper pulping (Krishnamachari et al. 1972; Luna 1996). *B. tulda* culms are used for construction, scaffolding, furniture, handicrafts and paper pulp while young shoots are pickled (Seethalakshmi and Kumar 1998). Being rich in phytosterols, *B. tulda* shoots can be used for production of sterol drugs (Srivastava 1990). However, both these bamboos exhibit extremely long flowering cycles of 25–40 years (Bahadur 1980; Dransfield and Widjaja 1995). Therefore, vegetative means of propagation need to be evolved to make the most of their potential. Rooting of culm/branch cuttings is the most suitable cloning option for propagation of bamboo species (McClure 1966; Troup 1992). However, the procedure has certain limitations; for instance, rooting recalcitrance and highly season-specific rooting behaviour of most bamboos. The different types of cuttings also behave differentially for induction of adventitious roots. Thus, we examined the influence of seasonal variation, indole-3-butyric acid (IBA) and cutting types on induction and growth of adventitious roots in *B. nutans* and *B. tulda* with the view of evolving efficient clonal multiplication procedures for both species.

Materials and methods

During four growing seasons i.e., Spring (S₁- mid February), Summer (S₂- mid May), Rainy (S₃- mid July) and Winter (S₄-

mid November) of 2006, mature culms of 12–15 nodes with lateral culm branches were collected from a five-year-old plantation. After excising of all the leaves single node culm cuttings (C_1) and culm-branch cuttings (C_2) were prepared. These cuttings were surface disinfected for 5 min with 0.25 (w/v) aqueous mercuric chloride solution and subsequently washed with sterilized water. Sixty sterilized cuttings of each type were immersed for 24 h in either water (T_1) or 2 mM indole-3-butyric acid (T_2). Each treatment consisted of three replicates each of 20 single node culm cuttings. The treated cuttings were horizontally placed and completely covered (10 cm deep) in sand beds of mist chamber maintaining $(70\pm 5)\%$ relative humidity and $(30\pm 2)^\circ\text{C}$ temperature.

After two months, the cuttings were scored for sprouting and adventitious rooting percentage, root number and root length. The data obtained were subjected to statistical analysis, employing analysis of variance (ANOVA), 'F'-test for significance at $p \leq 0.05$ and computing LSD values to separate means in different statistical groups using statistical software Assistat version 7.2 beta (Silva 2004).

Results and discussion

Bambusa nutans exhibited adventitious rooting in both types of cuttings in all planting seasons (Fig. 1a, b). However, better rooting success was recorded in culm cuttings compared to culm branch cuttings with the best rooting in the month of May. In contrast, culm cuttings of *B. tulda* recorded adventitious rooting only in February and May (Table 1; Fig. 1c). No root induction occurred in the culm branch cuttings. IBA treatment significantly enhanced rooting, root number and root length; registering 14 to 17 times improvement over control in the best rooting season.

Three factor-interaction (season \times cutting type \times IBA treatment) was significant for only rooting in *B. nutans* and all characteristics, except sprouting in *B. tulda*. Overall, the best rooting was recorded in IBA treated culm cuttings in the month of May 88% in *B. nutans* and 23% in *B. tulda* (Table 1).

Seasonal variation

Adventitious rhizogenesis in cuttings primarily depends on environment or internal factors, and/or their interactions. Season not only has a direct influence on the physiological state of the parent plant but also provides optimal conditions for induction and growth of adventitious roots in the detached cuttings. The importance of the physiological status of the bamboo plants in relation to stored nutrients (Gupta and Pattanath 1976), age (Dai 1981) and season (Agnihotri 1998; Singh et al. 2006) for subsequent adventitious rhizogenesis in culm cuttings has been reported. According to Banik (1987) the preparation of culm cuttings in the month of April–May was critical for obtaining adventitious rhizogenesis in many bamboos. In Indian conditions, good adventitious rooting in bamboo cuttings coincides with the cessation of winter and the onset of summer, resulting in emergence of new leaves/ side-branches, resumption of active extension (inter-

nodal) growth and upward mobilization of stored photosynthates from the underground rhizome (Agnihotri and Ansari 2000; Singh et al. 2002; 2006; Nain et al. 2007). In conformity, we recorded adventitious rhizogenesis only in the summer season in *B. tulda* and significantly superior root induction in *B. nutans* during summer season (May). This differential response may be attributed to the difference in periodical growth pattern of these bamboo species.

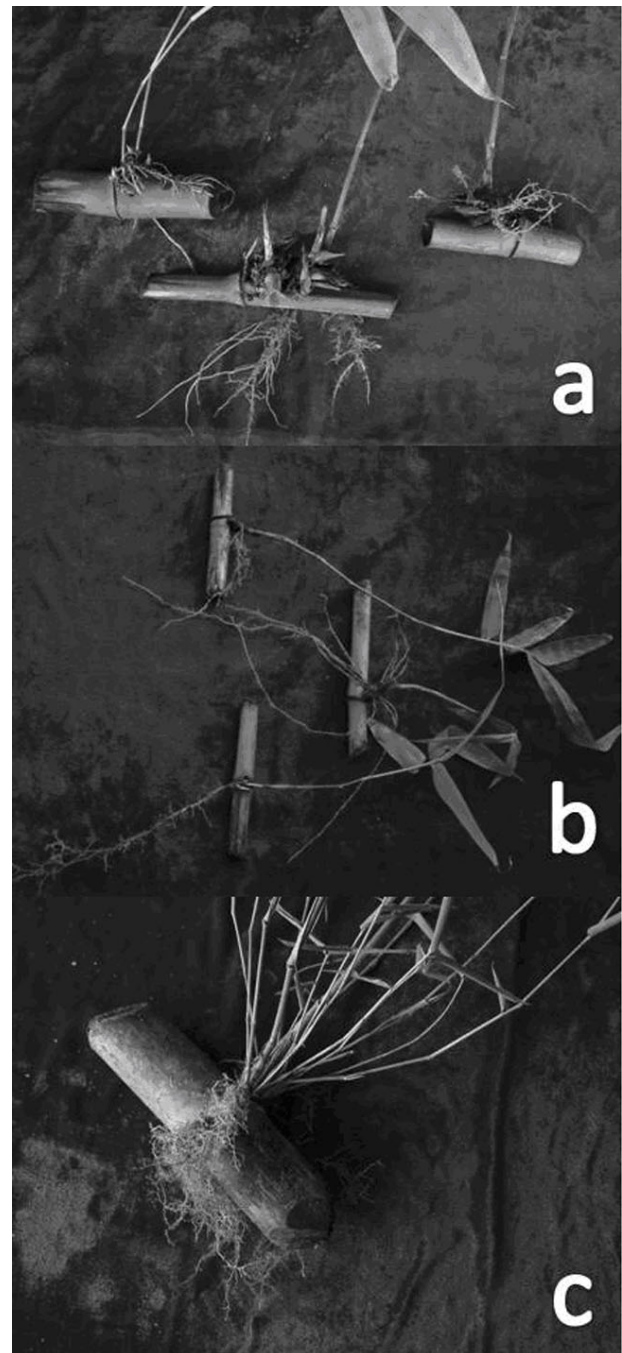


Fig. 1 Adventitious rooting in a. *B. nutans* culm cuttings; b. *B. nutans* culm-branch cuttings; and c. *B. tulda* culm cuttings

Table 1. The influence of season, cutting type and IBA treatment on adventitious rhizogenesis in *B. nutans* and *B. tulda*

Variables			Root characteristics							
			Sprouting (%)		Rooting (%)		Root number		Root length (cm)	
			<i>B. nutans</i>	<i>B. tulda</i>	<i>B. nutans</i>	<i>B. tulda</i>	<i>B. nutans</i>	<i>B. tulda</i>	<i>B. nutans</i>	<i>B. tulda</i>
Season	Cutting type	IBA treatment								
Spring (S1- mid February)	C ₁	T ₁	75	51.6	40	13.3	7.6	1.7	26.2	2.3
		T ₂	70	48.3	60	16.6	12.1	2.9	19.3	4
	C ₂	T ₁	58.3	10	48.3	0	7.8	0	30.8	0
		T ₂	40	13.3	61.6	0	7.3	0	34.9	0
Summer (S2- mid May)	C ₁	T ₁	70	18.3	81.6	1.6	8.5	0.5	36.1	0.6
		T ₂	63.3	21.6	88.3	23.3	14.2	5.8	44.4	8.5
	C ₂	T ₁	35	0	46.6	0	4.5	0	6.8	0
		T ₂	43.3	0	40	0	3.9	0	15.4	0
Rainy (S3- mid July)	C ₁	T ₁	50	31.6	41.1	0	3.5	0	6.4	0
		T ₂	65	30	55.6	0	4	0	7	0
	C ₂	T ₁	36.7	5	20	0	2.3	0	4.4	0
		T ₂	45	11.6	25.6	0	2.8	0	5.3	0
Winter (S4- mid November)	C ₁	T ₁	81.7	23.3	38.3	0	4.6	0	12.5	0
		T ₂	68.3	25	53.4	0	16.3	0	11.1	0
	C ₂	T ₁	25	0	15	0	1.9	0	5.3	0
		T ₂	10	1.6	21.8	0	8.5	0	7.8	0
LSD _{0.05}			NS	NS	16.57	6.83	NS	2.57	NS	2.02

Notes: C₁, single node culm cuttings; C₂, culm-branch cuttings

IBA treatment

Auxins, mainly IBA and NAA, have been employed with variable success in different types of bamboo cuttings e.g. *Bambusa vulgaris* (Uchimara 1978), *B. blumeana* and *Gigatochloa birs* (Bumalong and Tamolang 1980), *B. balcoa* (Seethalakshmi et al. 1989), *B. arundinacea* and *D. strictus* (Surendran et al. 1989), *B. vulgaris* and *D. strictus* (Agnihotri and Ansari 2000), *B. nutans* (Singh et al. 2002). Seemingly, exogenous auxin application elevates endogenous auxin level to the extent of inducing rooting in *B. tulda*, which is a very reluctant-to-root species (Kumar et al. 1990, 1994; McClure and Kennard 1955). In *B. nutans*, significantly superior proliferation of adventitious roots per cutting by the IBA treatment also underlines the need of auxin for the process. Thus, treatment of exogenous IBA helped adventitious root initiation in difficult-to-root *B. tulda* and enhanced their proliferation in relatively easy-rooting *B. nutans*.

Cutting type

Culm and culm-branches of bamboo possess variable capability for adventitious rhizogenesis due to their different ontogeny, which has been clearly manifested in the present investigation as culm cuttings exhibited markedly superior adventitious rooting than culm branch cuttings (Table 1). Earlier reports have also established superior root ability of culm cuttings compared to branch cuttings in *B. balcoa* (Surendran and Seethalakshmi 1985) and *Dendrocaamus asper* (Singh et al. 2004). Superior root induction in culm cuttings possibly relates to their juvenility status due

to proximal position and morphological features such as plump thick walls with abundant reserves of photosynthates. Joseph (1958) found high amount of starch in culm cuttings of *B. arundinacea* during good rooting months of February–March. Apparently, an optimal level of stored resources is needed for initiation, differentiation and growth of adventitious roots in cuttings.

In spite of low rooting response, culm-branch cutting produced in great number with very little economical value are good sources of propagation material, without damaging valuable culms. Use of culm-branch cuttings also reduces the weight and facilitates the transport of planting stock/propagules. The present study reveals that *B. nutans* may be propagated by culm cuttings in all seasons of the year and culm branch cuttings during the good rooting period i.e. February to May while *B. tulda* may be propagated by culm cuttings only.

Conclusion

Adventitious rhizogenesis was influenced much by season in *B. nutans* and by season, nature of cuttings and IBA treatment in *B. tulda*. Culm cuttings exhibited superiority over culm-branch cuttings for rooting in both species. However, rooting of culm-branch cuttings appeared to be a viable procedure for propagation in *B. nutans*. Thus, single node culm and culm-branch cuttings in *B. nutans* and culm cuttings in *B. tulda* treated with 2 mM IBA during February to May can be employed for their clonal multiplication.

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